I. Calculations

A. Using Absorbance at 260 nm to Measure DNA Concentration

A useful estimation of DNA concentration relates to the amount of oligonucleotide which, when dissolved in 1 mL of water, results in an absorbance of 1 when measured at 260 nm in a 1 cm path length cuvette. This is often simply called the A_{260} of a sample. The actual concentration can range from 39 µg/mL (for a homopolymer of C) to 20 µg/mL (for a homopolymer of A). For most practical experiments, an A_{260} of 1.0 represents approximately 33 mg of oligo with an equal mixture of the four bases.

\[ A_{260} \text{ conversion factors} \]

\[ A_{260} = 1.0 \quad \rightarrow \quad 33 \, \mu g/mL \, ssDNA \]
\[ \quad \rightarrow \quad 40 \, \mu g/mL \, ssRNA \]
\[ \quad \rightarrow \quad 50 \, \mu g/mL \, dsDNA \]

Molecular Weight of an Oligonucleotide

\[ MW_{\text{oligo}} = ((A \times 312.2) + (G \times 328.2) + (C \times 288.2) + (T \times 303.2) - 61) \]

where A,C,G,T represent the number of A's, C's, G's and T's in an oligo.

Molar conversions

1 µg of 1,000 bp DNA = 1.52 pmol (3.03 pmoles of ends)
1 µg of pBR322 DNA = 0.36 pmol DNA
1 pmol of 1,000 bp DNA = 0.66 µg
1 pmol of pBR322 DNA = 2.78 µg

Melting Temperature (Nucleic Acid Hybridization)

\[ T_m = \begin{cases} 
4^\circ C \, (G+C) + 2^\circ C \, (A+T) & \text{Up to 25 bp} \\
81.5^\circ C + 16.6 \, \log M + 0.41 \% (G+C) - 500 / n - 0.61 \% \text{ formamide} & \text{More than 25 bp}
\end{cases} \]

\[ M = [Na^+] \text{ in moles/liter} \; ; \; n = \text{length of shortest chain in duplex} \]

B. Other Information

Resuspension Buffers
1. Sterile Water (dd H₂O)
2. TE Buffer (10mM Tris-HCl, 1mM EDTA) pH 7.5

DNA Storage Conditions and Stability
Lyophilized (-20°C) = 6 months to several years
Lyophilized (25°C) = 2 months to 1 year
Dissolved (-20°C) = 1 month to 6 months
Dissolved (25°C) = 1 week to 3 months
DNA Conformations

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<tr>
<th>Helix type</th>
<th>Direction of rotation</th>
<th>Residues per turn</th>
<th>Rotation per residue</th>
<th>Helix rise per residue</th>
<th>Helix pitch</th>
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<tbody>
<tr>
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<tr>
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PAGE Purification Information

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<th>% Gel</th>
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<th>Xylene Cyanol</th>
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<td>8-60</td>
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References
2. George H. Keller, Mark M. Manak, DNA probes; p 15; M Stockton Press, '89.